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(71) Applicant: **SCOTIA HOLDINGS PLC**
Guildford Surrey GU1 1BA (GB)

(72) Inventor: **Horrobin, David Frederick, Dr.**
Guildford, Surray GU1 1BA (GB)

(74) Representative: **Sturt, Clifford Mark et al**
J. MILLER & CO.
34 Bedford Row,
Holborn
London WC1R 4JH (GB)

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(54) Use of a combination of arachidonic acid and dosohexaenoic acid in the preparation of e medicament for treating cell membrane abnormalities due to low C20 or C22 essential fatty acids

(57) The negative symptoms of schizophrenia and/or low cell membrane levels of EFAs may be treated with a combination of arachidonic acid and docosahexaenoic acid.

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Description

Essential fatty acids (EFAs) are essential constituents of all cell membranes including those in the brain. There are two series of EFAs, the n-6 derived from linoleic acid and the n-3 derived from alpha-linolenic acid. Their metabolic pathways are shown in Table 1.

TABLE 1

n-6		n-3
18:2 delta-9,12 (linoleic acid)		18:3 delta-9,12,15 (alpha-linolenic acid)
	↓ delta-6 desaturase	
18:3 delta-6,9,12 (gamma-linolenic acid)		18:4 delta-6,9,12,15 (stearidonic acid)
	↓ elongation	
20:3 delta-8, 11,14 (dihomo-gamma-linolenic acid)		20:4 delta-8,11,14,17
	↓ delta-5 desaturase	
20:4 delta-5,8,11,14 (arachidonic acid)		20:5 delta-5,8,11,14,17 ('eicosapentaenoic acid')
	↓ elongation	
22:4 delta-7,10,13,16 (adrenic acid)		22:5 delta-7,10,13,16,19
	↓ delta-4 desaturase	
22:5 delta-4,7,10,13,16		22:6 delta-4,7,10,13,16,19 ('docosahexaenoic acid')

Whereas in most cell membranes linoleic acid is an important component, this is not true of the brain. In the brain the two most important EFAs by concentration are arachidonic acid (AA) and docosahexaenoic acid (DHA). Dihomo-gamma-linolenic acid (DGLA) and eicosapentaenoic acid (EPA) are also important in the brain, but are present in much lower concentrations than AA and DHA. The other fatty acids of the n-3 and n-6 series are minor components but could nevertheless play important functional roles.

Schizophrenia is a common and serious psychiatric disorder affecting about 1 % of the population. It is diagnosed according to criteria set out in Diagnostic Standards and Methods III - Revised (DSM III R), a manual developed by the American Psychiatric Association and accepted in most countries. Patients classified as schizophrenic under DSM III

R may show a wide variety of signs and symptoms. There is discussion as to whether there are two or three separate sub-groups of patients within the overall DSM III R classification. Two of these sub-groups in particular, those with so-called "negative" and those with so-called "positive" symptoms have been much discussed. In the negative groups features of apathy, social withdrawal and a low level or absence of other symptoms predominate. The negative group may be clearly identified by using the negative component scores of standard psychiatric rating scales such as the Brief Psychiatric Rating Scale (BPRS) or by rating scales specifically designed to identify patients with the negative schizophrenia syndrome (sometimes also called the deficit syndrome). One very widely used negative scale is that devised by Andreasen et al. in Arch. Gen. Psychiatry (1982) Volume 39, pages 789-794, although others are available.

By the negative syndrome of schizophrenia herein is meant patients displaying at least one negative characteristic, identifiable by the rating scales described herein.

We have been interested for a number of years in the possibility that membrane abnormalities, particularly in the brain but also in other tissues, may play a role in schizophrenia. We have therefore performed analyses of the EFA levels in plasma and red blood cell phospholipids on various groups of schizophrenics and have compared those levels to those in normal individuals.

In plasma (Figure 1), all the EFAs showed a broadly unimodal distribution in both normal and schizophrenic individuals. Mean figures were different from normal in the schizophrenic patients, sometimes significantly so, and the scatter of values was often greater in the schizophrenics but there was no evidence of more than one schizophrenic population.

In the red blood cells (Figure 2) much the same was true for most of the fatty acids such as linoleic acid. Again distribution curves were often irregular in the schizophrenics as compared to the regular curves seen in the normals but for most fatty acids there was no clear evidence of more than one population. This was not true, however, of the two fatty acids particularly important in the brain, AA (20:4 n-6), and DHA (22:6 n-3), and of the other two fatty acids of lesser importance in the brain, EPA (20:5 n-3) and DGLA (20:3 n-6). For these fatty acids concentrations in red cell membranes showed a clear biphasic distribution of the kind illustrated for AA and DHA in Figs. 1 and 2. Further, one group of patients had values grouped around the normal region but another group of patients had a clearly different distribution with levels of the four fatty acids much lower than in normals or in other schizophrenics. These abnormal values appear related to the occurrence of symptoms generally regarded as being of the negative type.

This was confirmed in a study in which 13 patients with persistent negative schizophrenic symptoms were compared with 12 patients with persistent positive symptoms and 8 unequivocally normal individuals. In plasma, AA and DHA levels were relatively similar in all three groups. In contrast, in red cells, whereas the phospholipid DHA and AA levels were near normal in the positive schizophrenics, in the negative group they were highly significantly below normal ($p<0.001$ in each case). In the table below the figures are means ± SD. All figures are in mg/100 mg phospholipid

Red cell fatty acid	Normal (n=8)	Positive schizo (n=12)	Negative schizo (n=13)
AA	16.8 ± 0.5	15.5 ± 3.5	7.4 ± 2.3
DHA	5.1 ± 0.2	5.9 ± 1.8	2.2. ± 1.8

It appears therefore that around half of all patients currently classified as schizophrenic have a clear abnormality of EFA metabolism. These patients are those who exhibit the negative symptoms of the disease. Since plasma EFA levels are not far from normal, this group of patients clearly cannot incorporate the four major EFAs with 20 and 22-carbon atoms, namely AA, DHA, EPA and DGLA, into the red cell membranes normally. It is to be expected that the other, minor 20 and 22 carbon fatty acids show the same defect. It is also to be expected that this abnormality will be apparent in membranes throughout the body, particularly those of the brain. For fatty acids other than linoleic acid, there is a strong positive correlation between red cell and brain levels. This has been shown specifically for DNA by Connor in FASEB J, (1993) Volume 7, A152. Therefore, since neuronal membranes are critical for normal brain function, it is likely that these abnormalities are a cause of the negative schizophrenic symptoms in these patient groups.

The red cell membranes in patient groups with low 20 and 22-carbon fatty acids do contain some of these fatty acids. The mechanism for incorporation is therefore slow rather than absent. For this reason it will be possible to increase the amounts of the AA and DHA in cell membranes by supplying additional amounts of these fatty acids.

The present invention is therefore in one aspect to treat low cell membrane levels of C_{20} or C_{22} EFAs, by administering a combination of AA and DHA. It is preferred that the ratio of AA to DHA varies within the range of 20:1 to 1:20 preferably 5:1 to 1:5, and very preferably 3:1 to 1:3. Other EFAs such as EPA and DGLA may optionally be present. The EFAs may be administered in any appropriate form at a daily dose of from 10mg to 20g/day for each EFA, preferably 100mg to 10g/day, very preferably 200mg to 3g/day. Since gamma-linolenic acid (GLA) is rapidly converted to DGLA, it may replace DGLA in whole or in part in the formulation.

Another aspect of the invention is to treat all cell membrane abnormalities lying in low DHA and AA levels as above, by administering such EFA combinations.

A further aspect of the invention lies in the use of a combination of AA and DHA in the preparation of a medicament for the treatment of all cell membrane abnormalities as above.

The forms in which the fatty acids may be presented include any form which will allow the assimilation of the fatty acids into the body and into red cell membranes as indicated by a change in the composition of these red cell membranes on administration of the fatty acid. Suitable forms may be free fatty acids, physiologically acceptable esters of any type, mono-, di and triglycerides, amides, salts including sodium, potassium, lithium, calcium, zinc and other salts, and phospholipids including phosphatidyl (P) - choline, P-serine, P-inositol and P-ethanolamine and any other appropriate form. Routes of administration may include oral, enteral, parenteral (including subcutaneous, intramuscular, intravenous and intraperitoneal), topical, rectal, vaginal or by any other appropriate route.

As noted above, the schizophrenics with low red cell 20 and 22-carbon EFA levels appear on clinical examination to fall into the group with predominantly negative features. Some schizophrenics have both negative and positive features and it is not always possible to distinguish the group on clinical grounds. While, therefore, the treatment is particularly applicable to patients with any of the negative features of the disease, it can be applied to all schizophrenics in the knowledge that some will respond, while others who do not respond are most unlikely to be harmed, the fatty acids concerned being normal dietary components and bodily constituents. This, of course, is characteristic of the treatment of most diseases, where only some patients will respond to any particular measure.

The invention is further illustrated by the following Examples.

Examples

1. A triglyceride or a mix of triglycerides containing AA and DHA in a ratio ranging from 1:3 to 3:1 and formulated in a topical, oral or parenteral, rectal or vaginal dosage form such that a daily dose of between 20mg and 10g of each fatty acid can be delivered effectively. Such triglycerides might be a triglyceride with 2 molecules of DHA and one of AA, with 2 molecules of AA and one of DHA or with one of AA, one of DHA and one of another appropriate fatty acid formulated as 500mg or 1g soft gelatin capsules.

2. A mix of triglycerides as in 1, except that one triglyceride will contain 1-3 molecules of DHA and the other 1-3 molecules of AA, formulated as 500 mg soft gelatin capsules.

3. A triglyceride or mix or triglycerides as in 1 except that the fatty acids present are AA, DGLA, EPA and DHA.

4-6. As in 1-3 except that the fatty acids are present as free acids, for example a capsule containing 200mg AA, 200mg DHA and 200mg other carrier fatty acids.

7-9. As 1-3 except that the fatty acids are present as salts, such as lithium salts. The lithium salts, as well as being suitable for oral formulation as capsules or tablets with appropriate carriers, are also suitable for parenteral or enteral formulation, for example Li AA and Li DHA in a 20% alcoholic solution in sterile ampoules containing 500mg of each fatty acid for iv. administration.

10-12. As 1-3 except that the fatty acids are present as esters such as ethyl esters.

13-15. As 1-3 except that the fatty acids are present as amides.

16-18. As 1-3 except that the fatty acids are present as phospholipids such as P-choline, P-serine, P-inositol or P-ethanolamine or any other appropriate phospholipid.

Claims

1. Use of a combination of AA and DHA, optionally in association with other essential fatty acids, in particular GLA, DGLA and EPA, in the preparation of a medicament for the treatment of all cell membrane abnormalities lying in low C_{20} or C_{22} essential fatty acids (EFAs).

2. Use of a combination of AA and DHA, optionally in association with other essential fatty acids, in particular GLA, DGLA and EPA, in the preparation of a medicament for treating or correcting low cell membrane levels of C_{20} or C_{22} EFAs.

3. A use according to claim 1 or 2 wherein the medicament contains a weight ratio of n-6 to n-3 acids of 20:1 to 1:20, preferably 5:1 to 1:5 and more preferably 3:1 to 1:3.

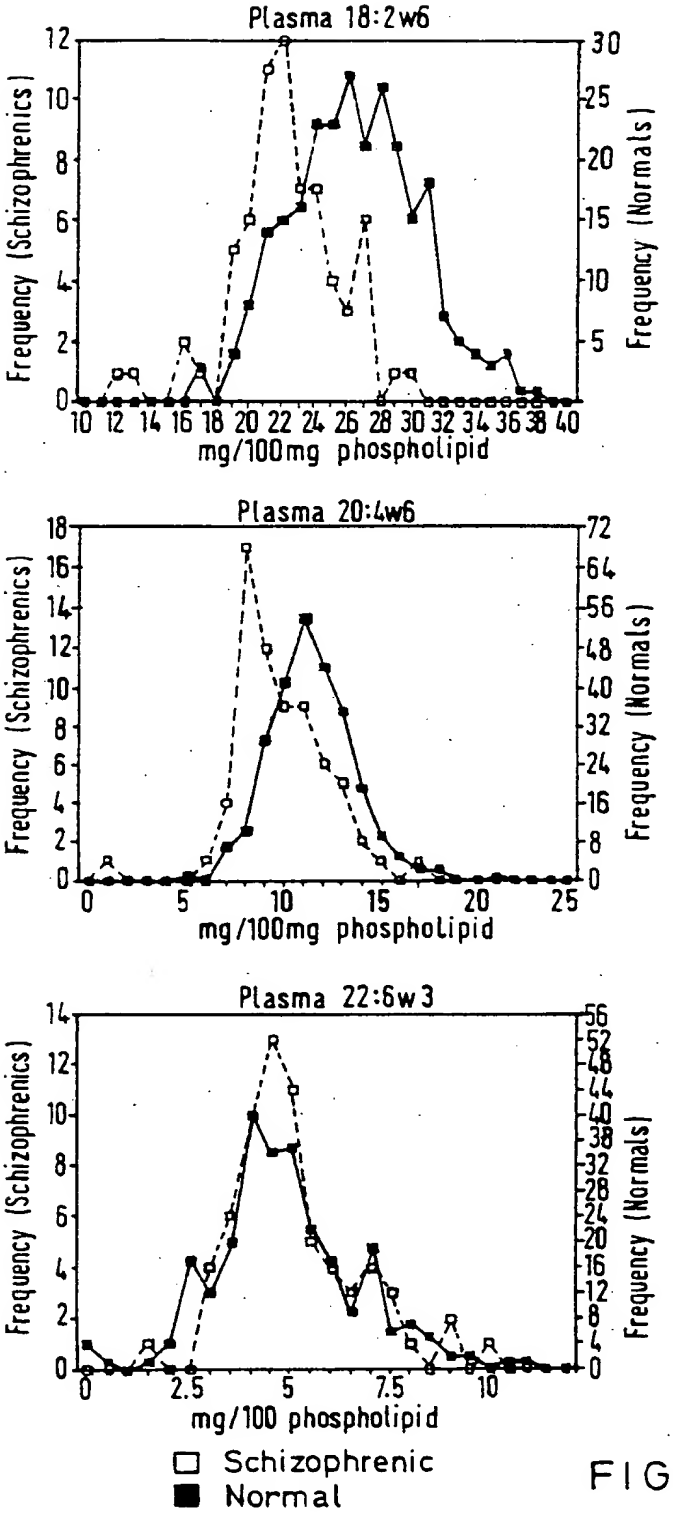
Plasma 18:2w6
Frequency (Schizophrenics)
Frequency (Normals)
mg/100mg phospholipid
Plasma 20:4w6
Frequency (Schizophrenics)
Frequency (Normals)
mg/100mg phospholipid
Plasma 22:6w3
Frequency (Schizophrenics)
Frequency (Normals)
mg/100 phospholipid
Schizophrenic
Normal

FIG.1

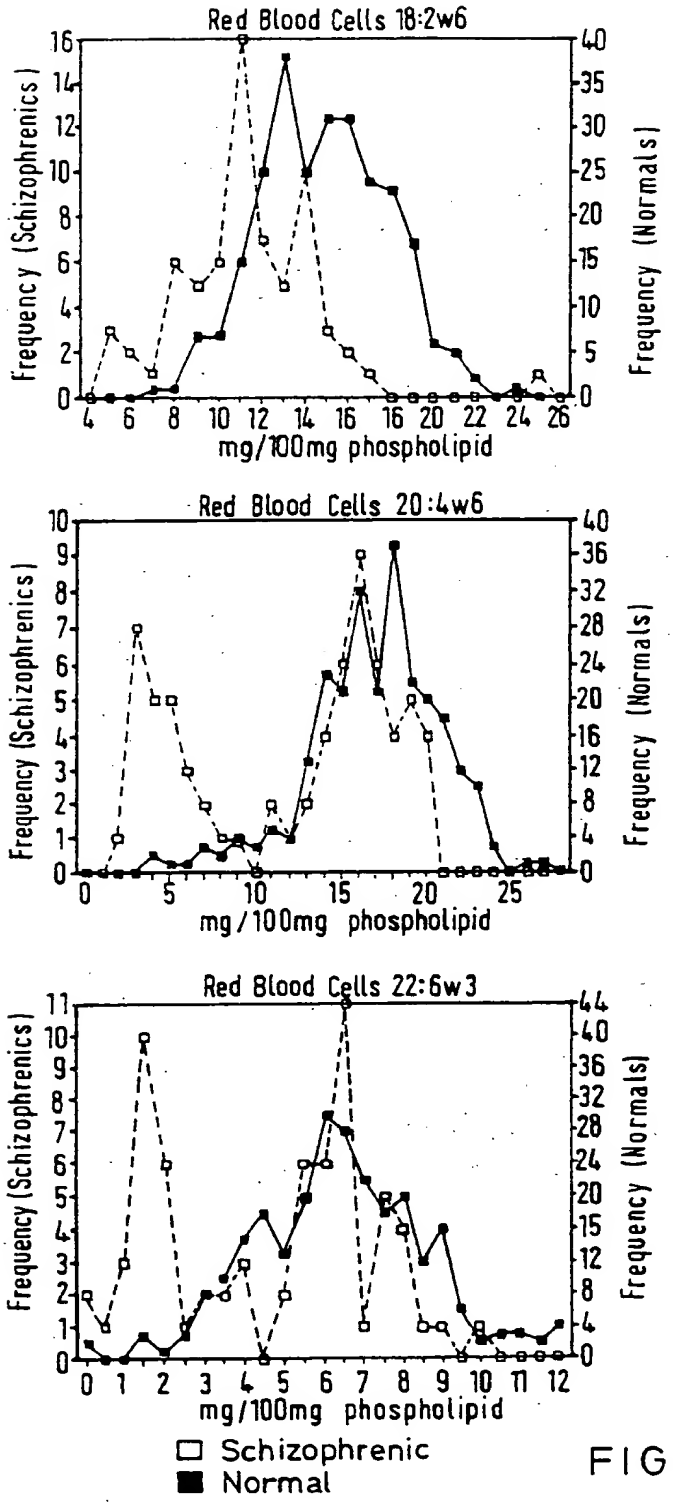
Red Blood Cells 18:2w6
Frequency (Schizophrenics)
Frequency (Normals)
mg/100mg phospholipid
Red Blood Cells 20:4w6
Frequency (Schizophrenics)
Frequency (Normals)
mg/100mg phospholipid
Red Blood Cells 22:6w3
Frequency (Schizophrenics)
Frequency (Normals)
mg/100mg phospholipid
Schizophrenic
Normal

FIG.2

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EUROPEAN SEARCH REPORT

Application Number
EP 96 10 5681

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.6)
X	EP-A-0 347 056 (EFAMOL HOLDINGS PLC) * page 4, line 46 - line 55; claims 6,7; example B4 *	1-3	A61K31/20 //(A61K31/20, 31:20)
X	EP-A-0 298 644 (EFAMOL HOLDINGS PLC) * page 3, line 36 - line 54 *	1-3	
X	EP-A-0 115 419 (EFAMOL LIMITED) * page 4, paragraph 2-3 *	1-3	
X	US-A-4 753 964 (HORROBIN) * the whole document *	1-3	
X,Y	NEUROSCIENCE AND BIOBEHAVIORAL REVIEWS, vol. 16, 1992, pages 193-205, XP002007080 P. WAINWRIGHT: "Do essential fatty acids play a role in brain and behavorial development?" * the whole document *	1-3	
X,Y	US-A-4 910 224 (HABIB NAGY A ET AL) 20 March 1990 * abstract; claim 1 *	1-3	TECHNICAL FIELDS SEARCHED (Int.Cl.6) A61K
X,Y	DE-A-40 17 979 (MEYER LUCAS GMBH & CO) 12 December 1991 * the whole document *	1-3	
X	EP-A-0 404 058 (MILUPA AG) 27 December 1990 * page 2 - page 3; claim 1; tables PAGE,5 *	1-3	

The present search report has been drawn up for all claims

Place of search	Date of completion of the search	Examiner
MUNICH	28 June 1996	Foerster, W

CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
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